

A within-species warning function for an aposematic signal

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Aposematic, or warning, signals are generally interspecific in form: one species advertises noxiousness to a predator or parasite species. In a study of the pipevine swallowtail butterfly (*Battus philenor*), we show that a pattern of colouration in the caterpillars that is considered to be aposematic in the context of attack by natural enemies also deters oviposition by conspecific females. In field and laboratory assays, females avoided oviposition on plants bearing live conspecific larvae. Females avoided oviposition on plants bearing artificially constructed models identical to larvae in shape, size and colour pattern. Finally, oviposition on plants harbouring a model bearing the larval colour pattern was reduced relative to plants bearing a leaf-green model, suggesting that the larval colour pattern was essential for avoidance. We discuss how intraspecific and interspecific processes might interact in the evolution of an aposematic signal.

Keywords: aposematism; warning colouration; oviposition; crypsis; predator avoidance; host-marking behaviour

1. INTRODUCTION

Aposematic signals promote avoidance of attack by predators and parasites, serving to 'warn' the natural enemy species of the signalling species' unprofitability. Despite the commonness of interspecific warning displays, their evolution has posed a problem to biologists (Mallet & Singer 1987; Endler 1988; Lindström *et al.* 1999, 2001; reviewed in Mallet & Joron 1999). Specifically, the first aposematic mutants to arise in a population of unpalatable individuals would seem to suffer increased detection by enemies while, at the same time, being too rare for enemies to learn to avoid. One proposition for circumventing these difficulties supposes that the initial costliness of a conspicuous pattern may be offset by a concurrent benefit unrelated to enemy avoidance (Endler 1988; Mallet & Joron 1999). While colour patterns in animals serve numerous functions, including communication, thermoregulation, water balance, prey acquisition, and crypsis (Endler 1988), relatively little effort has been expended to characterizing supplementary functions served by aposematic displays. An exception is assessment of the role of aposematic patterns in mating behaviour (Sherratt & Forbes 2001; Siddiqi *et al.* 2004).

In a study of the pipevine swallowtail butterfly (*Battus philenor* L.), we addressed the possibility that the larva's aposematic colour pattern, which is believed to warn natural enemies of its noxiousness, serves additionally to alert ovipositing conspecific females to the presence of a larva on a host plant. Specifically, we asked whether females discriminate against host plants bearing larvae and, if so, whether the effectiveness of such discrimination depends on the larval colour pattern.

2. MATERIAL AND METHODS

(a) Natural history and chemical ecology

The pipevine swallowtail belongs to the Tribe Troidini in the Family Papilionidae, a tribe whose members use only plants

within the Aristolochiaceae as hosts. Plants in this family contain aristolochic acids, which are biosynthetically related to benzoisoquinoline alkaloids (Chen & Zhu 1987; Fordyce 2000). Aristolochic acids are highly toxic to vertebrates and insects (Brower & Brower 1964; Chen & Zhu 1987; reviewed in Sime *et al.* 2000; Sime 2002). *B. philenor* larvae sequester aristolochic acids, and all life stages of the species contain them (Sime *et al.* 2000). Larvae offered to wolf spiders and paper wasps are refused (Bernays 1988; D. Papaj, personal observation). A generalist papilionid parasitoid (*Trogus* spp.) rejects *B. philenor* larvae and, owing to sequestered aristolochic acids, parasitoid offspring do not survive in them (Sime 2002). From a human standpoint, the butterfly is conspicuously coloured in egg, larval, and adult stages (photos in Electronic Appendix). Larvae feed during the day and rest in the open, as do members of other aposematic lepidopterous species.

In southern Arizona, *B. philenor* adults are common between late March and early September. The local host species, *Aristolochia watsoni*, is a small perennial, deciduous, recumbent vine with multiple stems which is abundant in washes and bordering areas. It varies in colouration from bright green to dark red. Larvae occur in a black form and a red form (photo in Electronic Appendix). The conspicuousness of each is enhanced by orange-tipped tubercles located medio-laterally along each side. The black form prevails across the butterfly's range; in southern Arizona, both forms occur, the red form, which develops under high temperatures, being more common.

(b) Field observations

In the field, we aimed to determine if females avoid laying eggs on plants bearing conspecific larvae. The site was 25 hectares of mesquite grassland on the University of Arizona Santa Rita Experimental Range (SRER) in Pima County, Arizona (31° 47.049' N; 110° 49.524' W). Between 09.00 and 16.00 h, we walked through the site until a female searching for hosts was detected. Host search is a highly stereotyped, readily identified behaviour in which females flutter near

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vegetation, making frequent turns and landing periodically. A female was observed either for 15 min search time (that is, time engaged in host search, exclusive of time spent nectaring, inspecting host plants, or laying eggs) or until she disappeared from view. When a female landed on a host plant, we recorded her behaviour (oviposition *versus* rejection). Oviposition is indicated by a stereotyped abdominal curling and ensuing quiescence. After a female left the plant, we recorded number of larvae, as well as the occurrence of newly laid eggs.

(c) *Laboratory assays: general methodology*

Wild females were collected from the SRER field site and allowed to oviposit on *Aristolochia fimbriata*. Larvae were raised on *A. fimbriata*, either on plants in gardens on the university campus or on cuttings of greenhouse-reared plants in the laboratory at ca 29 °C under 16 : 8 h light : dark photoperiod. Newly emerged butterflies were allowed to mate and lay eggs, and ensuing generations were reared from this stock, supplemented periodically with wild-collected material.

All assays were conducted in a 1.8 × 1.8 × 1.8 m³ nylon screen enclosure erected in the laboratory on a white linoleum floor and illuminated overhead by four 500-watt halogen fixtures suspended 2.14 m from the floor, supplemented by 20-watt fluorescent lighting mounted on the lab ceiling. We used green *A. watsoni* plants (photo in Electronic Appendix) in 10 cm diameter dark-green plastic pots. In experiments with live larvae, we used the black form which tends to develop under our comparatively cool laboratory and garden conditions. In experiments with larval mimics, we used one or both colour forms.

At the beginning of an assay, individually numbered, mated females were nectar-fed and allowed to search one at a time within the array. Ovipositions on plants were tallied by female according to treatment. Eggs were removed immediately after each oviposition. For each female, we attempted to record 10 ovipositions. In all experiments, pot position was rotated systematically throughout the day to control for location effects. In experiments employing models, models were rotated daily among plants, to control for variation among plants in attractiveness to females.

(d) *Laboratory assays: specific experiments*

Field observations cannot exclude the possibility that females avoided oviposition because plants bearing larvae were intrinsically different in some way from unoccupied plants, independent of the larvae themselves. In our first experiment, we therefore attempted to vary the presence of larvae independently of host characteristics. Six potted *A. watsoni* plants were paired according to size and growth, and distributed 40 cm apart in pairs on a cartesian grid on the screen cage floor. One randomly assigned member of each pair of plants received a third- or fourth-instar black-form larva, while the other was left untreated. Since an effect of larval presence might have been mediated by herbivore-induced changes in the plant, we ensured that three plants consistently received larvae over the course of the experiment and that three plants never received larvae.

Over the course of a day's assay, larvae fed little, if at all; plants changed little in size over time, relative to variation among plants in initial size. Nevertheless, females in this experiment may have responded to changes in the plants induced by feeding, or to some larval product such as faeces. This possibility was excluded in a second laboratory experiment by use of artificial models. Specifically, we sought

to determine whether ovipositing females discriminate against plants bearing visually mimicking models of red-form *B. philenor* larvae. Four *A. watsoni* plants were paired according to size and growth, and distributed 40 cm apart in a square. We placed a single artificial model of a red-form *B. philenor* larva (photo in Electronic Appendix) on two plants and left two plants untreated. Models consisted of synthetic plastic larval casts. A freshly killed 5th instar larva was immersed in pourable silicone mould-making rubber (Smooth-On, Inc.). Upon curing at ambient temperature, the mould was sliced open, larva removed, and hot glue injected into the mould. Upon cooling, the model was removed from the mould, and painted red with orange spots to mimic the red form of larvae. Paints (Golden Colors, Inc.) were matched to red and orange colours of wild-collected larvae using an S2000 spectrophotometer (Ocean Optics, Inc.) with tungsten-halogen light source, referenced to a white Spectralon standard (reflectance spectra in Electronic Appendix). Reflectance from larval surfaces in the ultraviolet (UV) was checked independently with the same spectrophotometer, using a deuterium source referenced against a Spectralon standard. UV reflectance of larva and paints relative to background was negligible and ignored in preparation of models. Models were allowed to dry for at least 2 days before use, then affixed with hot glue to green wire, and staked into the soil of a potted plant. The model was positioned such that it rested within the foliage as late-instar larvae on *A. watsoni* in the field usually do, i.e. in full, unobstructed view.

A third experiment extended the previous one to include visual mimics of the black form of *B. philenor* larvae (photo in Electronic Appendix), allowing us to compare efficacy of the two morphs. Models were constructed as described above. Six *A. watsoni* plants were divided into threes according to size and growth, and distributed 40 cm apart on a cartesian grid. We placed a red model on two plants, a black model on two plants, and left remaining plants untreated.

In a final laboratory experiment, we aimed to determine if deterrence of the visual mimics depended on their conspicuousness, by comparing the effect of the black-form model with that of a model painted green to match host leaves (photo in Electronic Appendix). Paint was matched to mature leaves from greenhouse-reared *A. watsoni* plants like those used in assays, employing the spectrophotometric protocols described above (reflectance spectra in Electronic Appendix). Six *A. watsoni* plants were paired according to size and growth characteristics, and distributed 40 cm apart in pairs on a cartesian grid. We placed a green model on two plants, a black model on two plants, and left remaining plants untreated.

(e) *Statistical analysis*

Matched pairs *t*-tests were performed. In assays with three treatments, we adjusted alpha levels in pair wise comparisons to 0.03 using Keppel's (1991) modified Bonferonni correction.

3. RESULTS

(a) *Field study*

In the field, host acceptance by females after landing was biased strongly against *A. watsoni* plants bearing larvae. Overall, 23% (71 of 307) of host plants found by 96 females bore larvae (median no. hosts found per female = 3, range = 1–8). Pooling across females, eggs were laid after landing on just 13% (9 of 71) of plants bearing larvae, compared to 47% (111 of 236) of plants lacking larvae.

In our analysis, we filtered the dataset to include only females that alighted both on plants with and plants without larvae, and analysed data with a matched pairs *t*-test. The pattern was nearly identical to that of pooled data. For 32 females remaining in the analysis, host acceptance after landing was consistently lower on plants bearing larvae (mean percentage plants accepted = 15%, s.e. = 4.0) than on plants lacking larvae (mean percentage plants accepted = 49%, s.e. = 4.0). The mean pair-wise difference in percentage acceptance of plants with *versus* without larvae was -33% (s.e. = 9.0; matched pairs *t*-test, $t_{31} = -3.81$, $p < 0.001$).

(b) Laboratory assays

Females deposited significantly fewer clutches on plants bearing live larvae than on plants bearing no larvae (figure 1a; matched pairs *t*-test, $t_{15} = 6.73$, $p < 0.0001$). Females also discriminated against plants with red larval models relative to control plants (figure 1b; matched pairs *t*-test, $t_{10} = 3.94$, $p < 0.002$). When mimics of both red and black forms were presented, females discriminated against both forms relative to control plants bearing no model (figure 2a; matched pairs *t*-tests; red form *versus* control: $t_{24} = 2.83$, $p < 0.005$; black form *versus* control: $t_{24} = 4.42$, $p < 0.0001$). The black form was somewhat more potent than the red form, but the trend is not statistically significant (figure 2a; matched pairs *t*-test, $t_{24} = 1.74$, $p < 0.1$). Because females rarely touched the models physically during plant inspection, females were most likely responding to visual cues.

Results of the final experiment which asked whether larval recognition depended on the *B. philenor* colour pattern, provided further support for the role of visual cues. In this experiment, plants bearing a visual mimic of the black form received significantly fewer ovipositions than control plants bearing no model or plants bearing a leaf-green model (figure 2b; matched pairs *t*-tests; black form *versus* control: $t_{47} = -2.94$, $p < 0.005$; black form *versus* green form: $t_{47} = -1.96$, $p < 0.03$). In contrast, plants bearing a model which was uniformly leaf-green, but otherwise identical in size and shape received only slightly fewer clutches on average, and the difference from control was not statistically significant (figure 2b; green form *versus* control; matched pairs *t*-test, $t_{47} = -1.24$, $p < 0.11$). These results suggest that the larva's colour pattern is of significance in deterring oviposition.

4. DISCUSSION

(a) Functional aspects of an intraspecific versus interspecific warning signal

An interspecific aposematic signal is presumed to benefit both signaller and receiver. A *B. philenor* larva signalling its noxiousness benefits by not being attacked by predator or parasitoid; would-be natural enemies benefit by not wasting time and energy, and not risking illness or death, in attacking the larva. Sherratt & Forbes (2001) extended the concept of aposematism to apply to intraspecific signals, proposing that bright colouration in male coenagrionid damselflies warns other males to stay away and avoid a wasteful interaction. Results presented here suggest that the *B. philenor* larval colour pattern deters oviposition by conspecific females. Might both signaller and receiver benefit from the

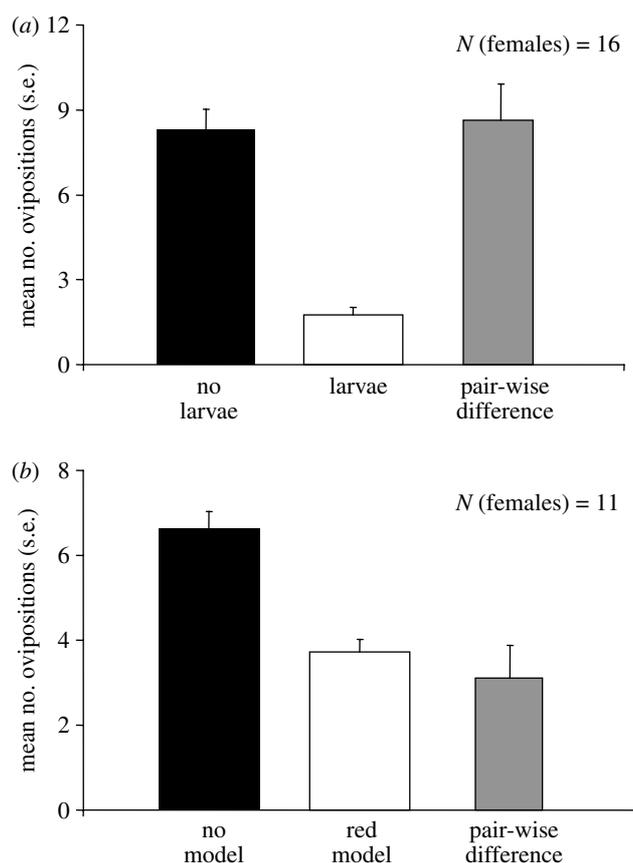


Figure 1. Mean number of ovipositions (+s.e.) on (a) plants bearing larvae *versus* unoccupied plants and; (b) plants bearing artificial model of red-form larvae *versus* unoccupied plants. Also shown is the mean pair-wise difference (+s.e.) in number of ovipositions in each experiment, the pair-wise difference being equal to the difference in number of ovipositions by a given test female across a pair of treatments.

deterrence, as in interspecific aposematic signals? The principal benefit of larval recognition is reduced competition among larvae. Competition may take multiple forms. First, a plant bearing more larvae will be consumed sooner and larvae will disperse at smaller sizes to find another plant; dispersal-related mortality in *B. philenor* is inversely proportional to size at dispersal (Rauscher 1980). Second, larvae may displace, injure, or even cannibalize eggs or young larvae. Third, larvae typically consume the best foliage on a plant first, leaving later-developing larvae to consume lower-quality foliage. Additionally, *Aristolochia* plants induce defences in response to herbivory (Fordyce 2003).

All four effects of larval occupation give incentive to adult females to avoid occupied plants, especially since offspring are not likely to compete successfully with older, competitively superior larvae. The incentive for the occupant(s) to signal their presence is less certain. Only the first and fourth effects of larval presence are likely to apply to signalling larvae and even these will depend on the difference in developmental stage between signaller and later-developing larvae. When the signalling larva is a fourth or fifth instar, for example, costs of competing with newly hatching larvae are probably negligible; however, for second or third instars, costs of competition, in terms of early dispersal, could be substantial. In entomophagous parasitoids, for

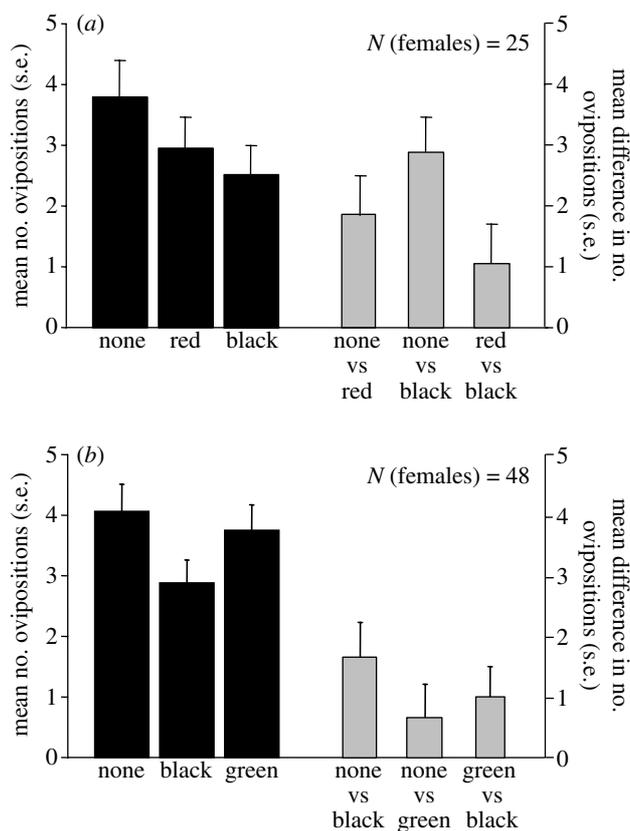


Figure 2. Mean number of ovipositions (+s.e.) on (a) plants bearing red-form model *versus* plants bearing black-form model *versus* unoccupied plants and; (b) plants bearing black-form model *versus* plants bearing cryptic green model *versus* unoccupied plants. Also shown is the mean pair-wise difference in number of ovipositions (+s.e.) for all possible treatment comparisons in each experiment, the pair-wise difference being equal to the difference in number of ovipositions by a given test female across a pair of treatments.

example, the first larvae to develop in a host are competitively superior, yet nevertheless experience reduced fitness if the host is ‘superparasitized’ (Visser *et al.* 1992).

(b) *Ramifications of dual function for the evolution of aposematism*

The present study demonstrates that a pattern of colouration in a butterfly larva considered to function as a warning of toxicity to natural enemies doubles as a warning to conspecific females of reduced offspring fitness. If the larval colour pattern evolved first or simultaneously in the context of larval recognition, this supplementary function could alleviate the problem of how an interspecific warning display spreads in a population (Endler 1988; see §1). However, the argument could work in reverse, since evolution of a signal of conspecific presence carries its own theoretical difficulties (reviewed in Nufio & Papaj 2001), which may be relieved by selection on the pattern’s interspecific function. Whether the *B. philenor* larval display evolved first in terms of larval recognition or first in terms of enemy avoidance, or simultaneously in both respects, is unclear at present. It would be useful to survey larval recognition within the troidines as well as sister tribes, in relation to the occurrence of larval noxiousness and warning displays.

(c) *Multiple receivers and active pattern elements*

The assertion that a given colour pattern is a signal both to conspecifics and to natural enemies suggests that natural selection might shape the pattern for detection by two or more species. Selection by multiple receivers could have several non-mutually exclusive outcomes. First, the pattern may consist of multiple features, each of which is adapted for a different receiver (Hebets & Papaj 2005). Alternatively, selection may result in a pattern whose features are co-adapted for all receivers. The latter possibility is not unreasonable, because the visual systems of animals as distantly related as insects and vertebrates show many convergent properties (Osorio *et al.* 1999). Swallowtail butterflies themselves have broad visual spectra, ranging from UV to red and perhaps near-infrared wavelengths, which probably encompass the collective spectra of possible natural enemies; their vision involves at least six distinct photoreceptor types, as well as screening pigments, which provide for sophisticated colour discrimination (Arikawa 2003).

Among candidate stimuli towards which butterfly and natural enemies might respond jointly is the unique shape of the larvae (photo in Electronic Appendix). In this case, the colour pattern might serve to make the shape stand out against foliage and soil. Another candidate is a colour common to both larval morphs, such as the orange in the spots that run along the mediolateral axis of the larva (photo in Electronic Appendix). Orange is an intriguing prospect, because it appears in three life stages (egg, larva, adult) and is also the colour of aristolochic acid, the source of noxiousness in *B. philenor*.

(d) *Conspicuousness versus uniqueness of the larval pattern*

The conspicuousness of an interspecific aposematic signal is considered to improve its efficacy, by enhancing detectability as well as the effectiveness with which natural enemies learn and remember the signal (Lindström *et al.* 1999; Speed 2000). Based on our results and what is known about papilionid vision, there is reason to think that the *B. philenor* larval pattern is conspicuous to adult females too. However, an alternative interpretation of our results is that females are responding selectively to a species-specific pattern. Distinguishing between these non-mutually exclusive possibilities might benefit by assaying conspicuous patterns other than the *B. philenor* pattern.

(e) *The role of non-visual larval recognition cues*

The low potency of visual mimics relative to live larvae suggests either that deviations between actual pattern and model pattern reduced female response or, alternatively, that other cues are involved. The latter possibility seems especially likely. Candidates include odours released by the damaged plant and odours associated with larval faeces (Hilker & Klein 1989). It is further possible that cues interact to improve signal efficacy and/or content (Hebets & Papaj 2005), perhaps through learning. *B. philenor* females readily learn visual and odour cues in association with host stimuli (Weiss & Papaj 2003).

(f) *Aggregation and dual warning function*

The potential for aggregations to enhance the effectiveness of interspecific aposematic signals has been a subject of

much debate (Gamberale & Tullberg 1998; Riipi *et al.* 2001; and citations within). Members of a given *B. philenor* clutch feed in aggregations in many locales; aggregations of first instars cause a short-term increase in host quality and promote larval growth (Fordyce 2003). In later instars, aggregations might not only enhance the warning towards natural enemies but might also promote larval recognition by ovipositing females.

We thank Heather Mallory, Laura Mojonier and Barrett Klein for technical assistance, and Michael Speed, Laura Carsten, Josh Ness, Jeff Oliver, Katy Prudic, Emilie Snell-Rood, Brad Worden, and Eileen Hebets and members of her lab group for comments. Work was funded by NSF-IBN grant no. 0112067.

REFERENCES

- Arikawa, K. 2003 Spectral organization of the eye of a butterfly, *Papilio*. *J. Comp. Physiol. A* **189**, 791–800. (doi:10.1007/s00359-003-0454-7.)
- Bernays, E. A. 1988 Host specificity in phytophagous insects: selection pressure from generalist predators. *Entomol. Exp. Appl.* **49**, 131–140. (doi:10.1007/BF00188246.)
- Brower, L. P. & Brower, J. V. Z. 1964 Birds, butterflies, and plant poisons: a study in ecological chemistry. *Zool. NY* **43**, 137–159.
- Chen, Z.-L. & Zhu, D.-Y. 1987 *Aristolochia* alkaloids. In *The alkaloids: chemistry and pharmacology*, vol. 31 (ed. A. Brossi), pp. 29–65. San Diego, CA: Academic Press.
- Endler, J. A. 1988 Frequency-dependent predation, crypsis and aposematic coloration. *Phil. Trans. R. Soc. B* **319**, 505–522.
- Fordyce, J. A. 2000 A model without a mimic: aristolochic acids from the California pipevine swallowtail, *Battus philenor hirsuta*, and its host-plant, *Aristolochia californica*. *J. Chem. Ecol.* **26**, 2567–2578. (doi:10.1023/A:1005588829864.)
- Fordyce, J. A. 2003 Aggregative feeding of pipevine swallowtail larvae enhances hostplant suitability. *Oecologia* **135**, 250–257.
- Gamberale, G. & Tullberg, B. S. 1998 Aposematism and gregariousness: the combined effect of group size and coloration on signal repellence. *Proc. R. Soc. B* **265**, 889–894. (doi:10.1098/rspb.1998.0374.)
- Hebets, E. A. & Papaj, D. R. 2005 Complex signal function: developing a framework of testable hypotheses. *Behav. Ecol. Sociobiol.* **57**, 197–214. (doi:10.1007/s00265-004-0865-7.)
- Hilker, M. & Klein, B. 1989 Investigation of oviposition deterrent in larval frass of *Spodoptera littoralis* (Boisd). *J. Chem. Ecol.* **15**, 929–938. (doi:10.1007/BF01015188.)
- Keppel, G. 1991 *Design and analysis*, 3rd edn. Englewood Cliffs, NJ: Prentice Hall.
- Lindström, L., Alatalo, R. V., Mappes, J., Riipi, M. & Vertainen, L. 1999 Can aposematic signals evolve by gradual change? *Nature* **397**, 249–251.
- Lindström, L., Alatalo, R. V., Lyytinen, A. & Mappes, J. 2001 Strong antiapostatic selection against novel rare aposematic prey. *Proc. Natl Acad. Sci. USA* **98**, 9181–9184. (doi:10.1073/pnas.161071598.)
- Mallet, J. & Joron, M. 1999 Evolution of diversity in warning color and mimicry: polymorphisms, shifting balance, and speciation. *Annu. Rev. Ecol. Syst.* **30**, 201–233. (doi:10.1146/annurev.ecolsys.30.1.201.)
- Mallet, J. & Singer, M. C. 1987 Individual selection, kin selection, and the shifting balance in the evolution of warning colours: the evidence from butterflies. *Biol. J. Linn. Soc.* **32**, 337–350.
- Nufio, C. R. & Papaj, D. R. 2001 Host marking behaviour in phytophagous insects and parasitoids. *Entomol. Exp. Appl.* **99**, 273–293. (doi:10.1023/A:1019204817341.)
- Osorio, D., Miklosi, A. & Gonda, Zs. 1999 Visual ecology and perception of coloration patterns by domestic chicks. *Evol. Ecol.* **13**, 673–689. (doi:10.1023/A:1011059715610.)
- Rausher, M. D. 1980 Host abundance, juvenile survival and oviposition preference in *Battus philenor*. *Evolution* **34**, 342–355.
- Riipi, M., Alatalo, R. V., Lindström, L. & Mappes, J. 2001 Multiple benefits cover detectability costs in aposematic aggregations. *Nature* **413**, 512–514. (doi:10.1038/35097061.)
- Sherrat, T. N. & Forbes, M. R. 2001 Sexual differences in coloration of Coenagrionid damselflies (Odonata): a case of intraspecific aposematism?. *Anim. Behav.* **62**, 653–660. (doi:10.1006/anbe.2001.1789.)
- Siddiqi, A., Cronin, T. W., Loew, E. R., Vorobyev, M. & Summers, K. 2004 Interspecific and intraspecific views of color signals in the strawberry poison frog *Dendrobates pumilio*. *J. Exp. Biol.* **207**, 2471–2485. (doi:10.1242/jeb.01047.)
- Sime, K. R. 2002 Chemical defense of *Battus philenor* larvae against attack by the parasitoid *Trogus pennator*. *Ecol. Entomol.* **27**, 337–345. (doi:10.1046/j.1365-2311.2002.00405.x.)
- Sime, K. R., Feeny, P. P. & Haribal, M. M. 2000 Sequestration of aristolochic acids by the pipevine swallowtail, *Battus philenor* (L.): evidence and ecological implications. *Chemoecology* **10**, 169–178.
- Speed, M. P. 2000 Warning signals, receiver psychology and predator memory. *Anim. Behav.* **60**, 269–278. (doi:10.1006/anbe.2000.1430.)
- Visser, M. E., Luyckx, B., Nell, H. W. & Boskamp, G. J. F. 1992 Adaptive superparasitism in solitary parasitoids—marking of parasitized hosts in relation to the pay-off from superparasitism. *Ecol. Entomol.* **17**, 76–82.
- Weiss, M. R. & Papaj, D. R. 2003 Colour learning in two behavioural contexts: how much can a butterfly keep in mind? *Anim. Behav.* **65**, 425–434. (doi:10.1006/anbe.2003.2084.)

The supplementary Electronic Appendix is available at <http://dx.doi.org/10.1098/rspb.2005.3186> or via <http://www.journals.royalsoc.ac.uk>.